RECENT RESEARCH ACTIVITIES

Coumarin-specific prenyltransferase, an enzyme crucial for furanocoumarin biosynthesis in parsley

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Furanocoumarins constitute a class of phenolic plant secondary metabolites, which are essentially found in four higher-plant families (Apiaceae, Rutaceae, Fabaceae and Moraceae). Furanocoumarins are important protectants for plants against pathogens or insects. In addition franocoumarins contribute to inter-specific competition by inhibiting the germination and growth of neighboring plants. In parsley (*Petroselinum crispum*), these compounds preferentially accumulate in oil ducts.

The metabolic profile of furanocoumarin differs largely depending on plant species, the age, and the tissues. There are two types of furanocoumarins derived from two parallel biosynthetic pathways, i.e., linear and angular furanocoumarins. It is hypothesized that these two pathways emerged from the result of co-evolution between plants and insects. Researches on interactions between butterfly larvae (*Depressaria pastinacella*) and an Apiaceae plant *Pastinaca sativa* showed that angular franocoumarins display a synergistic effect with linear ones, contributing the mixtures of linear and angular compounds more difficult for insects to detoxify. Thus, angular furanocoumarins have been hypothesized to appear later than linear ones over the course of plant evolution. This hypothesis is in line with the fact that angular furanocoumarins are always found concomitantly with linear structures in the plant kingdom.

The prenylation position of the common precursor of all furanocoumarins, umbelliferone, determines the type of furanocoumarin. Prenylation at C6 and C8 gives rise to the psoralen or angelicin derivatives, respectively. In this study, we identified a membrane-bound prenyltransferase PcPT from parsley, and characterized the enzymatic properties. PcPT expression in various parsley tissues is induced by UV irradiation, with a concomitant increase in furanocoumarin production *in planta*. PcPT has strict substrate specificity towards umbelliferone and dimethylallyl diphosphate, and a strong preference for the C6 position of the prenylated product (demethylsuberosin), which leads to the biosynthesis of linear furanocoumarins. In addition to the C6-prenylation, this enzyme also catalyzes the C8-prenylation in a much lesser extent. The PcPT protein was shown to be targeted to the plastids *in planta*. Metabolic engineering was performed in coumarin-producing plant *Ruta graveolens*, in which *PcPT* gene was over-expressed. It was found that PcPT expression in *Ruta graveolens* increased consumption of endogenous umbelliferone. Expression of PcPT together with a 4–coumaroyl CoA 2'–hydroxylase gene in *Nicotiana benthamiana*, which does not produce furanocoumarins, resulted in the formation of demethylsuberosin, indicative that furanocoumarin production may be reconstructed in non-producing plants. This finding suggested the plasticity of the furanocoumarin pathway, in particular for the emergence of angular structures during the course of plant evolution.

Acknowledgements

We are grateful to D. Baulcombe (University of Cambridge, UK) for pBIN61–P19, T. Nakagawa (Shimane University, Japan) for pGWB vectors, T. Mitsui (Niigata University, Japan) for WxTP–DsRed, H. Kouchi (National Institute of Agrobiological Sciences, Japan) for pHKN29 (GFP), and T. Duval (University of Lorraine, France) for technical assistance.

Reference

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